

AMENDMENTS TO THE CLAIMS

This listing of claims will replace all prior versions and listings of claims in the application:

LISTING OF CLAIMS:

1. (currently amended): A protein conjugate comprising i) a physiologically active polypeptide, ii) a non-peptidic polymer, and iii) an immunoglobulin, which are covalently linked to one another in that order, ~~and having~~

wherein the conjugate shows a prolonged in vivo half-life of the physiologically active polypeptide;

wherein the physiologically active polypeptide is selected from the group consisting of hormone, cytokine, enzyme, growth factor, transcription regulatory factor, blood factor, vaccine, structural protein, ligand protein, and receptor; and

wherein the immunoglobulin is selected from the group consisting of IgG, IgA, IgD, IgE, and IgM.

2. (Previously amended) The protein conjugate according to claim 1, wherein the non-peptidic polymer has two reactive groups at both ends, through which the non-peptidic polymer is covalently linked to the physiologically active polypeptide and the immunoglobulin of iii).

3. (Previously amended) The protein conjugate according to claim 2, wherein the immunoglobulin of iii) is covalently linked to at least two complexes of the physiologically active polypeptide and the non-peptidic polymer.

4. (canceled)

5. (currently amended): The protein conjugate according to claim [[4]]1, wherein the immunoglobulin of iii) is selected from the group consisting of IgG1, IgG2, IgG3, IgG4 and a mixture thereof.

6. (currently amended): The protein conjugate according to claim [[4]]1, wherein the immunoglobulin of iii) is a human immunoglobulin.

7. (Previously amended) The protein conjugate according to claim 1, wherein the immunoglobulin of iii) is selected from the group consisting of an immunoglobulin having the wild-type glycosylation, an immunoglobulin having an increased or decreased degree of glycosylation, an aglycosylated immunoglobulin and a combination thereof.

8. (Original) The protein conjugate according to claim 7, wherein the increase or decrease of the degree of glycosylation or aglycosylation of an immunoglobulin is conducted by a method selected from the group consisting of a chemical method, enzymatic method, biotechnological method and a combination thereof.

9. (Original) The protein conjugate according to claim 2, wherein the reactive group of the non-peptidic polymer is selected from the group consisting of aldehyde, propion aldehyde, butyl aldehyde, maleimide and succinimide derivative.

10. (Original) The protein conjugate according to claim 9, wherein the succinimide derivative is succinimidyl propionate, succinimidyl carboxymethyl, hydroxy succinimidyl or succinimidyl carbonate.

11. (Original) The protein conjugate according to claim 9, wherein the non-peptidic polymer has aldehyde groups at both ends.

12. (Previously amended) The protein conjugate according to claim 1, wherein the non-peptidic polymer is covalently linked at the ends thereof to the amino terminal, lysine residue, histidine residue or cysteine residue of the immunoglobulin and the amino terminal, lysine residue, histidine residue or cysteine residue of the physiologically active polypeptide, respectively.

13. (Original) The protein conjugate according to claim 1, wherein the non-peptidic polymer is selected from the group consisting of poly(ethylene glycol), poly(propylene glycol), ethylene glycol-propylene glycol copolymer, polyoxyethylated polyol, polyvinyl alcohol,

polysaccharide, dextran, polyvinyl ethyl ether, poly(lactic- glycolic acid), biodegradable polymer, lipid polymer, chitin, hyaluronic acids, and a mixture thereof.

14. (Original) The protein conjugate according to claim 13, wherein the non-peptidic polymer is poly(ethylene glycol).

15. (canceled).

16. (Currently amended) The protein conjugate according to claim ~~14~~¹⁵, wherein the physiologically active polypeptide is selected from the group consisting of human growth hormone, growth hormone releasing hormone, growth hormone releasing peptide, interferons, colony stimulating factor, interleukins, glucocerebrosidase, macrophage activating factor, macrophage peptide, B cell factor, T cell factor, protein A, suppressive factor of allergy, cell necrosis glycoprotein, immunotoxin, lymphotoxin, tumor necrosis factor, tumor inhibitory factor, transforming growth factor, alpha-1 antitrypsin, albumin, apolipoprotein-E, erythropoietin, hyper-glycosylated erythropoietin, factor VII, factor VIII, factor IX, plasminogen activator, urokinase, streptokinase, protein C, C-reactive protein, renin inhibitor, collagenase inhibitor, superoxide dismutase, platelet derived growth factor, epidermal growth factor, osteogenic growth factor, osteogenesis stimulating protein, calcitonin, insulin, atriopeptin, cartilage inducing factor, connective tissue activator protein, follicle stimulating hormone, luteinizing hormone, FSH releasing hormone, nerve growth factor, parathyroid hormone, relaxin, secretin, somatomedin, insulinlike growth factor, adrenocorticotrophic hormone, glucagon,

cholecystokinin, pancreatic polypeptide, gastrin releasing peptide, corticotropin releasing factor, thyroid stimulating hormone, receptor, receptor antagonist, cell surface antigen, ~~monoclonal~~ antibody, polyclonal antibody, antibody fragment including Fab, Fab', F(ab')₂, Fd and scFv, and virus-derived vaccine antigen.

17. (Original) The protein conjugate according to claim 16, wherein the physiologically active polypeptide is human growth hormone, interferon alpha, interferon beta, granulocyte colony stimulating factor or erythropoietin.

18. (Currently amended) A method for preparing the protein conjugate of claim 1, comprising:

(a) covalently linking at least one physiologically active polypeptide and one ~~whole~~ immunoglobulin with at least one non-peptidic polymer having reactive groups at both ends, wherein the molar ratio of the immunoglobulin to the non-peptidic polymer ranges from 1: 5 to 1: 10; and

(b) isolating a protein conjugate comprising essentially the active polypeptide, the immunoglobulin and the non-peptidic polymer, which are interlinked covalently,

wherein the physiologically active polypeptide is selected from the group consisting of hormone, cytokine, enzyme, growth factor, transcription regulatory factor, blood factor, vaccine, structural protein, ligand protein, and receptor; and

wherein the immunoglobulin is selected from the group consisting of IgG, IgA, IgD, IgE, and IgM.

19. (Previously amended) The method according to claim 18, wherein step (a) comprises:

(a1) covalently coupling one end of the non-peptidic polymer with either the immunoglobulin or the physiologically active polypeptide;

(a2) isolating from the resulting reaction mixture a complex comprising the non-peptidic polymer coupled with the immunoglobulin or the physiologically active polypeptide; and

(a3) covalently coupling the free end of the non-peptidic polymer of the complex with the immunoglobulin or physiologically active polypeptide, to produce a protein conjugate comprising the physiologically active polypeptide, the non-peptidic polymer and the immunoglobulin, which are covalently interlinked.

20. (Original) The method according to claim 19, wherein the molar ratio of the physiologically active polypeptide to the non-peptidic polymer in step (a1) ranges from 1: 2.5 to 1: 5.

21. (Canceled).

22. (Previously amended) The method according to claim 19, wherein the molar ratio of the complex obtained in step (a2) to the physiologically active polypeptide or immunoglobulin in step (a3) ranges from 1: 1 to 1: 3.

23. (Original) The method according to claim 19, wherein steps (a1) and (a3) are performed in the presence of a reducing agent.

24. (Original) The method according to claim 23, wherein the reducing agent is sodium cyanoborohydride, sodium borohydride, dimethylamine borate or pyridine borate.

Claims 25-53 (Cancelled).

54. (New) The protein conjugate according to claim 1, wherein the immunoglobulin is IgG.

55. (New) The protein conjugate according to claim 1, wherein the physiologically active polypeptide is selected from the group consisting of human growth hormone, interferon alpha, interferon beta, granulocyte colony stimulating factor and erythropoietin, and wherein the immunoglobulin is IgG.

56. (New) The protein conjugate according to claim 1, wherein the non-peptidic polymer is poly(ethylene glycol) and the immunoglobulin is IgG.

57. (New) The protein conjugate according to claim 56, wherein the physiologically active polypeptide is selected from the group consisting of human growth hormone, interferon alpha, interferon beta, granulocyte colony stimulating factor, and erythropoietin.